

Omega-3 long-chain PUFA intake during pregnancy and allergic disease outcomes in the offspring: a systematic review and meta-analysis of observational studies and randomized controlled trials¹

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ABSTRACT

Background: There is some evidence that increased maternal intake of omega-3 (n-3) long-chain polyunsaturated fatty acids (LC-PUFAs) during pregnancy may reduce the incidence of immunoglobulin E (IgE)-mediated allergic disease.

Objective: We aimed to evaluate prenatal n-3 LC-PUFA dietary exposure in observational studies and n-3 LC-PUFA supplementation in randomized controlled trials (RCTs) on outcomes of IgE-mediated allergic disease.

Design: We conducted searches of the Cochrane Central Register of Controlled Trials, PubMed, Ovid MEDLINE, EMBASE, CINAHL, SCOPUS, and Web of Science to 30 July 2015. We included prospective cohort studies that showed an association between maternal fish or n-3 LC-PUFA intake during pregnancy and RCTs with a prenatal intervention to modify maternal n-3 LC-PUFA intake and outcomes of allergic disease (eczema, rhino-conjunctivitis, asthma) or sensitization in the offspring.

Results: A total of 13 publications from 10 prospective cohort studies and 7 publications representing 5 unique RCTs were included. Three RCTs were combined in a meta-analysis for selected outcomes. Nine of 13 observational study publications and 5 of 7 publications from RCTs found a protective association between increased prenatal n-3 LC-PUFA or fish intake and incidence of allergic disease symptoms in the child. Meta-analysis was limited because of the heterogeneity of the RCTs. Pooled results showed a significant reduction in the incidence of "atopic eczema," "any positive SPT [skin-prick test]," "sensitization to egg," and "sensitization to any food" in the first 12 mo of life [RRs (95% CIs): 0.53 (0.35, 0.81), $P = 0.004$; 0.68 (0.52–0.89), $P = 0.006$; 0.55 (0.39–0.76), $P = 0.0004$; and 0.59 (0.46, 0.76), $P < 0.0001$, respectively].

Conclusions: Our systematic review and meta-analysis was suggestive of benefits of increased n-3 LC-PUFAs in the maternal diet and outcomes of childhood allergic disease. However, due to the inconsistency in results, the hypothesis linking maternal n-3 LC-PUFA intake to childhood allergic disease cannot unequivocally be confirmed or rejected.

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Keywords: allergic disease, n-3 LC-PUFA, pregnancy allergy prevention, fatty acids, randomized controlled trial, prenatal supplementation, asthma, eczema, hayfever

INTRODUCTION

There is general consensus that the worldwide prevalence of allergic disease has escalated too rapidly to be attributed to genetic changes alone. Although the cause may have mixed etiology, it is widely accepted that the current allergy epidemic is attributable to a changing environment, including lifestyle factors and diet. Dietary ratios of n-6 to n-3 PUFAs have changed from an equal balance of n-6:n-3 (1:1) to almost 30:1 (n-6:n-3) in some Western cultures (1). These profound changes in the type of fat consumed parallel the increasing prevalence of atopy and allergic disease (atopic eczema, IgE-mediated rhino-conjunctivitis, IgE-mediated allergic asthma), posing the hypothesis that this imbalance may have a causal relation. When consumed, n-3 and n-6 PUFAs compete for the same enzymes required for conversion into their long-chain (LC)⁷ derivatives (LC-PUFAs). Diets high in n-6 PUFAs due to the increased consumption of linoleic acid (18:2n-6)-rich vegetable oils and arachidonic acid (20:4n-6) from meat result in a predominance of proinflammatory arachidonic acid in tissues. This leads to biochemical and physiologic changes consistent with a greater propensity to an inflammatory allergic response (2). Plausible mechanisms therefore exist whereby diets high in n-3 LC-PUFAs may modulate the development of IgE-mediated allergic disease and regulate immune responses. Data from clinical and animal studies suggest that dietary n-3 LC-PUFAs in early life may influence immune system development and immune cell function,

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⁷ Abbreviations used: DHQ, diet history questionnaire; DIPPE, Diabetes Prediction and Prevention; FFQ, food-frequency questionnaire; ISAAC, International Study of Asthma and Allergies in Childhood; LC, long-chain; RCT, randomized controlled trial; SPT, skin-prick test.

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reducing inflammatory responses (3); however, clinically beneficial effects are more conflicting.

Initiating events of allergic disease occur early in immune development, with antigen-specific reactivity detected in cord blood at birth and as early as 23 wk of gestation in the fetus (4). Hence, it is our hypothesis that there may be a window of opportunity to modulate the fetal immune system before it has been programmed to an allergic phenotype by increased maternal supply of anti-inflammatory n-3 LC-PUFAs (5).

Published reviews on the effect of n-3 LC-PUFAs in the primary prevention of allergic disease have been conducted; however, not all are systematic and most do not include observational studies (5–9). Two reviews pooled results of n-3 LC-PUFA exposure during fetal life, infancy, and childhood, increasing the variability of results and potentially diluting any causal effect (5, 7). In the complex field of nutrition and health outcomes, multiple levels of research evidence should be considered when establishing causal effects (10). We therefore included epidemiologic studies observing n-3 PUFA/LC-PUFA dietary exposures and randomized controlled trials (RCTs) with an n-3 LC-PUFA intervention and subsequent associations or effects on IgE-mediated allergic disease. By limiting this systematic review to exposures or interventions that commenced in the intrapartum period, we aim to develop a clearer understanding of the effect to the developing fetus, before commencement of the progression of atopy (“atopic march”) and establishment of allergic disease symptoms.

METHODS

Our systematic review is reported according to the PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses) guidelines (11). We included all prospective studies, including longitudinal observational studies and RCTs. Observational studies were included if they examined an association between maternal fish or n-3 LC-PUFA intake during pregnancy and clinical outcomes of allergic disease or sensitization in the offspring. RCTs and quasi-randomized trials that evaluated an intervention modifying maternal n-3 LC-PUFA intake during pregnancy with a parallel control group or placebo on the clinical outcomes of allergic disease (eczema, rhino-conjunctivitis, asthma) or sensitization in the offspring were eligible for inclusion in the review. Animal studies, cross-sectional studies, and retrospective and case-control studies were excluded. All of the included participants were pregnant women, regardless of gestation, and their offspring. There was no restriction placed on the atopic predisposition of the women, their offspring, or the age of follow-up. Included studies were required to report maternal intake of n-3 LC-PUFAs via dietary intake or supplements during the pregnancy period. Studies of maternal n-3 LC-PUFA consumption or supplementation in the postnatal period only (breast-feeding or direct supplementation of the infant) were excluded.

The primary outcome measure of this review was the incidence of atopic disease (i.e., IgE-mediated allergic disease) or sensitization in the offspring during infancy, childhood, or adolescence. The presence of IgE-mediated allergic disease is defined as a clinician diagnosis, parent report of symptoms of allergic disease, or parent report of a physician’s diagnosis. Sensitization is defined as a positive skin-prick test (SPT) or IgE serology indicating sensitization. Studies that reported immune biomarkers

by laboratory assessment in the absence of evaluation of symptoms or clinical diagnosis of allergic disease in the offspring were excluded.

A comprehensive search for publications was undertaken using the following databases from inception to 30 July 2015: Cochrane Central Register of Controlled Trials (CENTRAL issue 6, 2012; <http://www.cochranelibrary.com>), PubMed (1966 to July 2015; <http://www.ncbi.nlm.nih.gov>), Ovid MEDLINE (1946 to July 2015; <http://ovidsp.tx.ovid.com>), EMBASE (1974 to July 2015; <https://www.embase.com>), CINAHL (<https://www.ebscohost.com/nursing/products/cinahl-databases/cinahl-complete>), SCOPUS (<http://www.scopus.com>), and Web of Science (<http://login.webofknowledge.com>). Databases were searched for relevant publications by using a search strategy tailored for each database on the basis of the following PubMed search terms: (Fatty Acids, Omega-3[mh:noexp] OR Omega 3 Fatty Acid*[tw] OR n-3 PUFA[tw] OR n-3 Fatty Acid*[tw] OR n-3 Polyunsaturated Fatty Acid*[tw] OR Docosahexaenoic Acid*[tw]) OR fish[tw] AND (Pregnancy[mh] OR Pregnant*[tw] OR Perinatal[tw] OR Prenatal[tw] OR Antenatal[tw] OR maternal[tw] OR Gestation[tw]) AND (child[mh] OR child*[tw] AND offspring[tw] OR Infant[mh] OR Infant*[tw] OR Adolescen*[tw] OR Youth*[tw]) AND (Asthma[tw] OR wheez*[tw] OR respiratory*[tw] OR IgE-Mediated Hypersensitiv*[tw] OR Immediate hypersensitiv*[tw] OR Atopic Hypersensitiv*[tw] OR Atopy[tw] OR Type I Hypersensitiv*[tw] OR Allergic rhinitis[tw] OR Hay fever[tw] OR Hayfever[tw] OR Atopic Eczema[tw] OR Atopic Dermatitis[tw] OR Allerg*[tw]).

We supplemented our search by cross-checking the reference lists of relevant retrieved publications identified by the search and recent review articles. No date restrictions were imposed, although results were limited to human studies. The titles and abstracts of all articles retrieved by the search were reviewed by one author (KPB) to assess eligibility for inclusion in the review. If there was insufficient information in the abstract to warrant exclusion of an article, the full text of the article was retrieved to determine eligibility. Any uncertainty with regard to the inclusion of publications was resolved after discussion with a second reviewer (MM).

Data relating to dietary exposure or interventions, outcomes, potential effect confounders, and study quality were extracted by use of a standardized data extraction form. Extracted information included characteristics of study participants, type of exposure measure (cohort), maternal exposure to/supplementation of fish or n-3 LC-PUFAs, timing and length of intervention, and type and timing of outcome measures. Studies included in this review varied markedly in terms of the methods, timing, and type of intervention and timing and type of reported outcome measures. We therefore reported study results for all observational studies and a number of RCTs descriptively, including their applicability and limitations. Meta-analysis was considered only for RCTs with comparable timing of outcome assessment (age of offspring). Outcomes of allergic disease that met this criterion include the following: “atopic eczema,” “sensitization to egg,” “sensitization to any food,” and “any sensitization.” Other outcomes, including asthma symptoms and rhino-conjunctivitis, were unable to be combined due to heterogeneity with definition of outcomes between studies and the young age of follow-up (2 and 3 y). Preplanned analysis of allergic disease outcomes were grouped according to the age of the offspring at the time of outcome assessment. Sensitivity analysis was conducted to

investigate the influence of a single study on the overall estimate; $P < 0.05$ was considered to be significant for all tests. All analyses were performed by using Review Manager Software 5.3 (Copenhagen: The Nordic Cochrane Centre, The Cochrane Collaboration), and heterogeneity was assessed by comparing the CIs of the results of individual studies and with the I^2 statistic. When there was an absence of significant heterogeneity, the results were pooled by using a fixed-effects model. When substantial heterogeneity was detected ($I^2 > 50\%$), possible causes were explored, and a random-effects model was used. Continuous outcomes are reported as mean differences with 95% CIs.

RESULTS

The search returned a total of 1133 publications. After removal of duplicates, 543 publications remained. Of these, 478 publications were excluded because they did not meet the predefined inclusion criteria. A total of 65 full-text articles were reviewed for eligibility, resulting in 45 exclusions for reasons listed in **Figure 1**. Thirteen publications from 10 unique prospective cohort studies and 7 publications representing 5 unique RCTs were included in the review. Three trials with comparable timing of outcome assessments for “any sensitization,” “sensitization to egg,” and “sensitization to any food” and 2 trials with atopic eczema outcomes at 12 mo were combined in the meta-analysis.

Participants

All of the studies were conducted in industrialized countries. Seven of the observational studies enrolled healthy pregnant women presenting at antenatal or obstetric care clinics between 5 and 39 wk of gestation, irrespective of atopy status (12–18). One study from The Netherlands selectively enrolled participants depending on their nationality (19) and another restricted enrollment to Danish-speaking participants (20). Two remaining studies enrolled full-term infants, collecting maternal diet information retrospectively (21–24). One of these studies (with 3 publications included in this review) was a nested nutrition follow-up of the Diabetes Prediction and Prevention (DIPP) study in Finland (22–24). Mothers were enrolled after birth and confirmation that their child was at risk of developing diabetes by the presence of human leukocyte antigen in cord blood. Characteristics and results of observational studies are presented in **Table 1**.

All of the included RCTs recruited women during the antenatal period from clinical care settings. Four of the 5 RCTs enrolled women who were at risk of delivering a fetus with atopic disease (i.e., first-degree relative with history of allergic disease) (25–29); however, only one of these trials confirmed atopy in the mother by SPT before enrollment (30). The remaining trial included women attending their routine 30-wk midwife clinic visit, regardless of atopy status (31). Characteristics and results of RCTs are presented in **Table 2**.

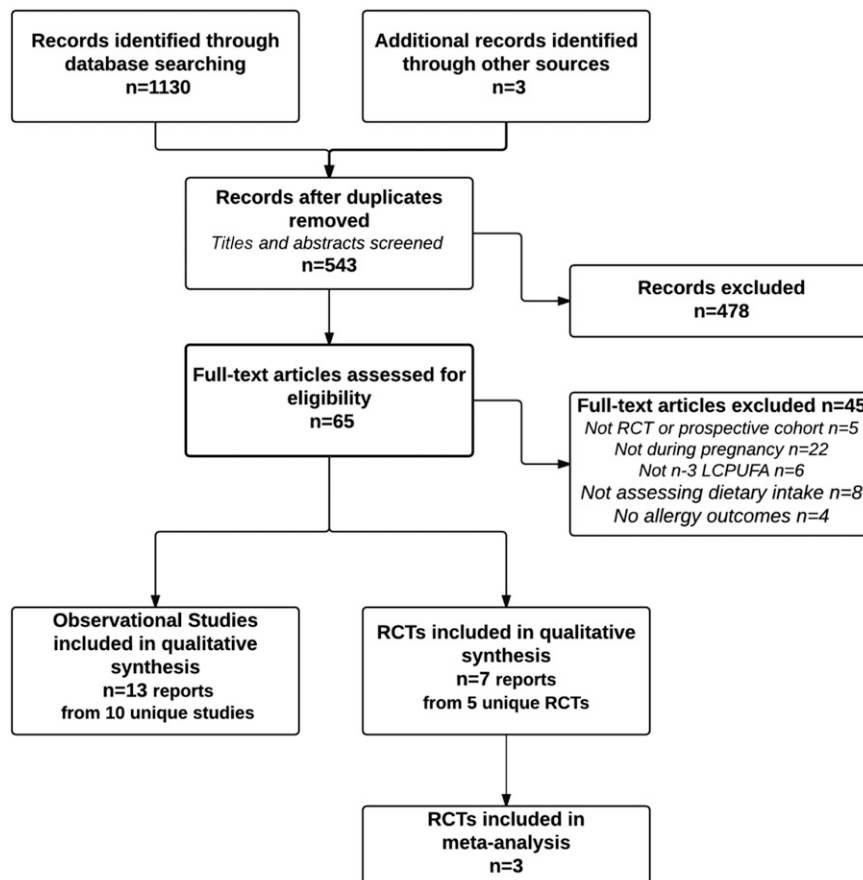


FIGURE 1 PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses) flow diagram depicting the literature search and progress of observational studies and RCTs identified and included in the systematic review and meta-analysis. LC-PUFA, long-chain PUFA; RCT, randomized controlled trial.

TABLE 1
Observational studies of maternal n-3 LC-PUFA intake during pregnancy and allergic disease in the offspring¹

Authors, year (ref)	Participants	Exposure assessment	Maternal exposure	Outcome assessment	Results	Adjusted variables
Maslova et al., 2013 (20)	n = 28,936 Danish-speaking women only from Danish National Birth Cohort; Denmark, 1996–2002	360-Item FFQ at 25 wk gestation (diet past 4 wk) and fish intake via telephone interview, ×2	Frequency of fish intake: zero intake; monthly or less; more than monthly; weekly, low frequency; weekly, high frequency Mean fish or LC-PUFA intake = NR	Parent report of doctor-diagnosed asthma, wheeze symptoms, and wheezing episodes (n) at 18 mo	Zero vs. high-frequency maternal fish intake (by phone interview) associated with an increased risk of child asthma diagnosis (OR: 1.30; 95% CI: 1.05, 1.63; P = 0.02); assessment of fish intake by FFQ showed no association	Parental education, occupation, maternal age, parity, prepregnancy BMI, smoking and exercise during pregnancy, gestational weight gain, BF duration, BW, GA, sex, parental history of asthma and allergies
	Same population as (20)	360-Item FFQ at 25 wk gestation (diet past 4 wk) and fish intake via telephone interview, ×2	Frequency of fish intake: zero intake; monthly or less; more than monthly; weekly, low frequency; weekly, high frequency Mean fish or LC-PUFA intake = NR	Parent report of doctor-diagnosed asthma and hay fever; parent report of asthma and allergic rhinitis symptoms (ISAAC); National Patient Registry and Register of Medicinal Product Statistics for asthma at 7 y	Zero vs. high-frequency maternal fish intake (by phone interview) associated with an increased risk of “ever admitted asthma” (OR: 1.46; 95% CI: 0.99, 2.13; P = 0.05) and “ever prescribed asthma” (OR: 1.37; 95% CI: 1.10, 1.71; P = 0.01); no association with wheeze, recurrent wheeze, or allergic rhinitis	Parental education, occupation, maternal age, parity, prepregnancy BMI, smoking and exercise during pregnancy, gestational weight gain, BF duration, BW, GA, sex, parental history of asthma and allergies
Miyake et al., 2013 (18)	n = 1354; Kyushu Okinawa Maternal and Child Health Study; Japan, 2007–2008	150-Item DHQ, 5–39 wk gestation (diet past 4 wk)	Median fish intake: Q1, 23.4 g/d; Q2, 37.5 g/d; Q3, 49.8 g/d; Q4, 71.3 g/d Mean intake fish = 46.9 g/d; n-3 PUFAs = 2.3 g/d	Parent report of symptoms of wheeze and eczema by ISAAC at 23–29 mo	No significant association between maternal fish intake and wheeze or eczema; maternal intake of EPA and EPA plus DHA associated with reduced risk of wheeze (OR: 0.73; 95% CI: 0.50, 1.04; P-trend = 0.02; OR: 0.70; 95% CI: 0.49, 1.00; P-trend = 0.02, respectively).	Maternal age; gestation; region; parity; maternal and paternal education, asthma, atopic eczema, and rhinoconjunctivitis; maternal smoking in pregnancy; family income; BW; sex; household smoking during first year; BF duration

(Continued)

TABLE 1 (Continued)

Authors, year (ref)	Participants	Exposure assessment	Maternal exposure	Outcome assessment	Results	Adjusted variables
Leermakers et al., 2013 (19)	n = 2796 Dutch-only women from Generation-R study recruited at antenatal visit; Netherlands, 2002–2006	293-Item semiquantitative FFQ; 10–21 wk gestation (diet past 3 mo)	Total, lean, and fatty fish consumption; median fish intake = 83 g/wk; median fatty fish intake = 32 g/wk	Parent report of wheezing and doctor-attended eczema by adapted ISAAC at 1, 2, 3, and 4 y	No association of maternal total fish consumption with childhood wheezing at 1–4 y; maternal consumption of 35–69 g fatty fish/wk vs. no fatty fish increased the overall risk of eczema (OR: 1.17; 95% CI: 1.00, 1.38); maternal consumption of ≥70 g fatty fish/wk was not associated with eczema (OR: 1.06; 95% CI: 0.88, 1.38); no association with maternal total or lean fish consumption and childhood eczema	Maternal age, parity, SES, asthma or atopy, vegetable intake, periconception folic acid, education, psychological distress, prepregnancy BMI, smoking and alcohol consumption during pregnancy, pets, BW, GA, sex, BF, daycare, lower respiratory tract infections, complementary feeding
Nwaru et al., 2012 (23)	n = 2441 Infants from the Finnish Type 1 DIPP Study; Finland, 1997–2004	181-Item FFQ at delivery, retrieved at 3 mo postpartum (diet in eighth month of pregnancy)	Total n–3 PUFA intake: Q1, <5.55 g/d; Q2 and Q3, 5.55–7.34 g/d; Q4, 7.35–11.28 g/d Total n–3 PUFA from fish: Q1, <0.38 g/d; Q2 and Q3, 0.38–0.98 g/d; Q4, 0.99–5.50 g/d Mean n–3 PUFAs = 3 • 2 g/d	Parent report of rhino- conjunctivitis, wheeze, and atopic eczema by modified ISAAC at 5 y	Higher ratio of n–6:n–3 fatty acids during pregnancy associated with an increased risk of rhino- conjunctivitis in the offspring by 5 y of age (OR: 1.37; 95% CI: 1.07, 1.77); no association with increased total n–3 PUFAs, n–3 PUFAs (from fish or plants), DHA, or EPA and eczema, rhinitis, or wheeze	Hospital of birth, GA, maternal age, education, smoking during pregnancy, parental asthma and rhino- conjunctivitis, delivery, sex, siblings, pets at home by 1 y of age, vitamin C, zinc, selenium, vitamins D and E
Lumia et al., 2011 (22)	n = 2679; Same population as (23)	181-Item FFQ at delivery, retrieved at 3 mo postpartum (diet in eighth month of pregnancy)	Total n–3 PUFA intake: Q1, <2.24 g/d; Q2 and Q3, 2.24–3.84 g/d; Q4, >3.84 g/d Total fish intake: Q1, <10.67 g/d; Q2 and Q3, 10.67– 31.99 g/d; Q4, >31.99 g/d Mean fish products = 21 g/d	Parent report of asthma by modified ISAAC combined with antiasthmatic medication data from Finnish Social Insurance Institution at 5 y	Low maternal intake of ALA and low total n–3 PUFA intake associated with an increased risk of asthma in the offspring (OR: 1.67; 95% CI: 1.12, 2.48; OR: 1.66; 95% CI: 1.11, 2.48; P = 0.036); no association between maternal intake of oily fish and fish products and the risk of asthma in the offspring	Maternal age, education, parity, mode of delivery, GA, BW, sex, area of birth, maternal smoking, parental asthma or rhino- conjunctivitis, pets, farming, contact with cow stable during the first year of life, BF duration

(Continued)

TABLE 1 (Continued)

Authors, year (ref)	Participants	Exposure assessment	Maternal exposure	Outcome assessment	Results	Adjusted variables
Nwaru et al., 2011 (24)	<i>n</i> = 931; Same population as (23)	181-item FFQ at delivery, retrieved at 3 mo postpartum (diet in month 8 of pregnancy)	<i>n</i> -3 PUFA quartile data = NR Mean fish products = 23 g/d	Total and specific IgE analysis (egg, cow milk, fish, wheat, HDM, cat, timothy grass, and birch) at 5 y	No significant association with <i>n</i> -3 PUFA intake and sensitization	Place and season of birth, maternal age, smoking during pregnancy, education, parental asthma and rhino-conjunctivitis, siblings, sex, GA at birth
Jedrychowski et al., 2011 (17)	<i>n</i> = 469 Women recruited from ambulatory prenatal clinics; New York and Poland, 2001–2004	FFQ in second and third trimester (food items or intake timing NR)	Frequency of smoked, fried, roasted, and grilled fish: T1, ≤90 g/wk; T2, 91–205 g/wk; T3 >205 g/wk Mean fish intake = 148 g/wk	Face-to-face interview with regard to infant health and physician-confirmed infantile eczema at 3, 6, 9, and 12 mo	Maternal fish intake of >205 g/wk associated with a reduction in risk of eczema by 43% (OR: 0.57; 95% CI: 0.35, 0.93; <i>P</i> = 0.047)	Maternal age, education, atopy, duration BF, siblings, and damp or moldy house
Miyake et al., 2009 (16)	<i>n</i> = 763 Women recruited from obstetric clinics to the Osaka Maternal and Child Health Study; Japan, 2001–2003	150-item DHQ, any stage of pregnancy; mean: 17.7 wk gestation (diet past 4 wk)	Total <i>n</i> -3 PUFA intake: Q1, 1.7 g/d; Q2, 2.2 g/d; Q3, 2.5 g/d; Q4, 3.0 g/d Total fish intake: Q1, 23.4 g/d; Q2, 38.7 g/d; Q3, 51.7 g/d; Q4, 73.2 g/d Mean <i>n</i> -3 PUFAs = 2.4 g/d; mean fish = 48.4 g/d	Parent-reported symptoms of wheeze and eczema by ISAAC at 16–24 mo	High maternal ALA intake associated with a reduced risk of wheeze in the offspring (OR: 0.52; 95% CI: 0.28, 0.97), although the inverse exposure-response relation was not significant; no association with eczema.	Maternal age, GA, location, family income, maternal and paternal education, asthma, atopic eczema and allergic rhinitis, maternal intake of vitamins D and E, changes in diet during previous month, smoking, siblings, sex, BW, household smoking, BF
Willers et al., 2008 (15)	<i>n</i> = 2832 Women recruited from antenatal clinic; Netherlands, 1997–1999	Dietary questionnaire self-completed between 30th and 36th week of gestation (diet past 4 wk)	Frequency of fish intake: rarely (never to 1–3 times/mo); regularly (1 to >4 times/wk; daily (≥1/d)	Parent report of asthma symptoms by ISAAC at 1–8 y	No overall association between maternal fish consumption during pregnancy and childhood wheeze, dyspnea, steroid use, or asthma symptoms	Child sex, maternal education, smoking, parental allergy, smoking in the home at 8 y, BF, siblings, BW, maternal overweight 1 y after pregnancy, maternal supplements in pregnancy, region, study arm
Chatzi et al., 2008 (14)	<i>n</i> = 468 Women presenting for antenatal care at general practices in Menorca, Spain, 1997–1998	96-item FFQ by face-to-face interview 3 mo postpartum (referred to pregnancy period)	Frequency of fish intake: never; times per year; monthly; weekly (≥1 time/wk, 85.8%); other categories NR	Annual questionnaire by interviewer; 1–6 y; SPT (HDM ×2, grass pollen, olive tree, mixed graminace, parietaria) at 6.5 y	Maternal fish intake of ≥2.5 times/wk inversely associated with persistent wheeze (OR: 0.34; 95% CI: 0.13, 0.84; <i>P</i> ≤ 0.05)	Maternal age, atopy, social class and education, maternal supplement use and smoking, maternal and paternal asthma, BF, lower track respiratory infections at 1 y, BW, birth order, GA, number of siblings, and BMI at 6.5 y

(Continued)

TABLE 1 (Continued)

Authors, year (ref)	Participants	Exposure assessment	Maternal exposure	Outcome assessment	Results	Adjusted variables
Ronieu et al., 2007 (12)	<i>n</i> = 468; Same population as (14)	35-Item FFQ by face-to-face interview 3 mo postpartum (referred to pregnancy period)	Frequency of fish intake: never; times per year; monthly; weekly (≥ 1 time/wk; 85.8%); other categories = NR	Annual questionnaire by interviewer 1–6 y. Specific IgE (HDM $\times 2$ and mixed grass pollens) at 4 y. SPT (HDM $\times 2$, grass pollen, olive tree, mixed Gramineae, Parietaria) at 6 y	Fish intake 2.5 times/wk vs. once was protective against risk of eczema (OR: 0.73; 95% CI: 0.55, 0.98; <i>P</i> = 0.03); no significant association between fish intake and IgE concentrations at age 4 y; fish intake 2.5 times/wk vs. once was protective against positive SPT to HDM and atopic wheeze (OR: 0.68; 95% CI: 0.46, 1.01; <i>P</i> = 0.05; OR: 0.55; 95% CI: 0.31, 0.96; <i>P</i> = 0.03 respectively)	Maternal age, smoking, and pre-pregnancy BMI; maternal and paternal atopy, asthma, social class; child sex; GA; BW; parity; BF; pets; child BMI at age 6.5 y; child's fish intake
Willers et al., 2007 (13)	<i>n</i> = 1212 Women recruited from antenatal clinic; United Kingdom, 1997–1999	150-Item semiquantitative FFQ mailed to women at 32 wk gestation; diet past 2–3 mo	Total fish intake: never (<i>n</i> = 107); 1 time/wk (<i>n</i> = 255); ≥ 1 time/wk (<i>n</i> = 831) Total oily fish: never (<i>n</i> = 629); 1 time/wk (<i>n</i> = 414); ≥ 1 time/wk (<i>n</i> = 161) Mean intakes = NR	ISAAC spirometry, bronchodilator response, exhaled nitric oxide, and SPT (cat, timothy grass, egg, and HDM) at 5 y	Maternal fish consumption ≥ 1 time/wk vs. never associated with a decreased risk of doctor-confirmed eczema, and doctor-confirmed hay fever (OR: 0.57; 95% CI: 0.35, 0.92; <i>P</i> -trend = 0.08; OR: 0.58; 95% CI: 0.32, 1.06; <i>P</i> -trend = 0.028; OR: 0.28; 95% CI: 0.06, 1.19; <i>P</i> -trend = 0.04; respectively); no consistent association between maternal fish consumption and atopic sensitization, spirometry, bronchodilator response, or exhaled nitric oxide	Maternal age, education, smoking during pregnancy, paternal social class, maternal asthma and atopy, child's BW, sex, presence of older siblings, BF, smoking in the child's home at 5 y
Sausenthaler et al., 2007 (21)	<i>n</i> = 2641 Healthy full-term infants from maternity hospitals in birth cohort study (LISA); Germany, 1997–1999	FFQ administered after birth (median: 3 d) (diet past 4 wk)	High vs. low fish intake: < 2 times/mo or never to ≥ 4 times/wk (individual categories = NR); high intake (T1, <i>n</i> = 122); low intake (T2 and T3, <i>n</i> = 322) Mean intakes = NR	Parental report of doctor-diagnosed eczema at 6, 12, 18, and 24 mo; total and specific IgE analysis (egg, cow milk, wheat, peanut, soybean, codfish, HDM, cockroach, cat dander, mixed molds, seasonal allergens) at 2 y	High vs. low maternal fish intake inversely associated with doctor-diagnosed eczema (OR: 0.75; 95% CI: 0.57, 0.98; <i>P</i> ≤ 0.05); no significant association between fish consumption and allergic sensitization	Location, maternal age at delivery, maternal smoking, parental education, exclusive BF for 4 mo, parental history of atopic diseases, season of birth, child sex

¹ ALA, α -linolenic acid; BF, breastfeeding; BW, birth weight; DHQ, diet history questionnaire; DIPP, Diabetes Prediction and Prevention; FFQ, food-frequency questionnaire; GA, gestational age; HDM, house dust mite; ISAAC, International Study of Asthma and Allergies in Childhood; LC-PUFA, long-chain PUFA; LISA, influence of lifestyle factors on the development of the immune system and allergies; NR, not reported; Q, quartile; ref, reference; SES, socioeconomic status; SPT, skin-prick test; T, tertile.

TABLE 2
RCTs of maternal n-3 LC-PUFA supplementation during pregnancy and allergic disease in the offspring¹

Authors, country, year (ref)	Setting and participants	Intervention and timing	Outcomes	Follow-up (age; completed/enrolled, n; rate, %)	Results
Dunstan et al., Australia, 2003 (30)	n = 98 Atopic, nonsmoking pregnant women recruited from antenatal clinic, 1999–2001	Fish-oil capsules, 4×1 g/d; 3700 mg n-3 LC-PUFAs (56.0% DHA, 27.7% EPA); control: olive oil capsules 20 wk GA until delivery	Clinical examination and history by physician to determine incidence of asthma, atopic eczema, and food allergy and SPT (hen's egg, cow milk, peanut, HDM, and cat)	12 mo; 83/98; 85	Sensitization to egg at 12 mo lower in the intervention group (OR: 0.34; 95% CI: 0.11, 1.02; $P = 0.05$); no difference in frequency of eczema; however, the intervention group had less severe disease (OR: 0.09; 95% CI: 0.0, 0.94; $P = 0.045$); recurrent wheeze, persistent cough, and diagnosed asthma all lower in intervention group (10/40 vs. 12/43, 5/40 vs. 11/43, and 2/40 vs. 6/43, respectively; NS)
Olsen et al., Denmark, 2008 (31)	n = 533 Women attending midwife clinic at routine 30-wk assessment, 1989–1990	Fish-oil capsules, 4×1 g/d; 2700 mg n-3 LC-PUFAs (23% DHA, 32% EPA); control 1: olive oil capsules; control 2: no supplement 30 wk GA until delivery	Asthma-related diagnosis extracted from the Danish National Patient Registry	16 y; 528/533; 99	Lower incidence of diagnosis of “any asthma” in the fish-oil group (2/263 vs. 8/136; $P = 0.03$); lower incidence of diagnosis of “allergic asthma” in the fish-oil group (8/263 vs. 11/136; $P = 0.01$)
Furuhjelm et al., Sweden, 2009 (26)	n = 145 Women with fetus at high risk of allergic disease, recruited from antenatal clinic or local newspapers, 2003–2005	Fish-oil capsules, 9×0.5 g/d, 2700 mg n-3 LC-PUFAs (1.6 g EPA, 1.1 g DHA); control: soy oil capsules; 25 wk GA until 3.5 mo postnatally	Clinical examination by nurse (pediatrician exam if food allergy or eczema) at 3, 6, and 12 mo; SPT (cow milk, egg, and wheat); IgE antibodies to egg, milk, and wheat at 3 and 12 mo; SPT (cow milk, egg, and wheat)	6 mo: 117/145; 81; 12 mo: 115/145; 79	Incidence of IgE-associated eczema at 0–6 mo was lower in the intervention group, although not significant [4/52 (8%) vs. 13/65; $P = 0.06$]; incidence of IgE-associated eczema, sensitization to egg, and period prevalence of “any positive SPT” was lower in the intervention group at 0–12 mo (4/52 vs. 15/63, $P = 0.02$; 6/52 vs. 16/63, $P = 0.02$; 8/52 vs. 20/63, $P = 0.04$; respectively)
Furuhjelm et al., Sweden, 2011 (25)	n = 145; Same population as (26)	—	Clinical examination by pediatrician and SPT (cow milk, egg and wheat, cat, timothy, and birch)	24 mo; 143/145; 98	Cumulative incidence (0–24 mo) of any IgE-mediated disease, positive SPT to egg, any positive SPT, and any IgE-associated eczema was lower in the intervention group (6/54 vs. 19/62, $P = 0.01$; 7/52 vs. 18/61, $P = 0.04$; 10/52 vs. 22/61, $P = 0.048$; 5/54 vs. 15/63, $P = 0.04$; respectively); no difference between groups for “any asthma,” IgE-associated conjunctivitis, IgE-associated rhinoconjunctivitis at 24 mo, or cumulative incidence at 0–24 mo

(Continued)

TABLE 2 (Continued)

Authors, country, year (ref)	Setting and participants	Intervention and timing	Outcomes	Follow-up (age; completed/enrolled, n; rate, %)	Results
Noakes et al., United Kingdom, 2012 (27)	n = 123 Women with low habitual oily fish intake with a fetus at high risk of atopy recruited from antenatal clinic; year NR	2 × 150 g salmon portions/wk (1160 mg DHA, 570 mg EPA); control: habitual diet low in oily fish; 20 wk GA until delivery	Total serum IgE from cord blood at birth, symptom diary cards, examination by nurse, and SPT (HDM, cat, dog, tree mix, grass mix, egg, salmon, cow milk)	Birth: 107/123; 87; 6 mo: 86/123; 70	No significant difference in total IgE at birth between groups; no significant difference in total IgE, SPT, incidence of eczema, severity of eczema, or wheeze between groups
Palmer et al., Australia, 2012 (29)	n = 706 Women with a fetus at high risk of atopy recruited from antenatal clinics, 2005–2007	Fish-oil capsules 3 × 0.5 g/d; 900 mg n-3 LC-PUFAs (800 mg DHA, 100 mg EPA); control: vegetable oil capsules; 21 wk GA until delivery	Clinical examination by physician to determine incidence of asthma, eczema, and food allergy SPT (cow milk, egg, wheat, tuna, peanut, grass pollen, perennial ryegrass, olive tree pollen, <i>Alternaria tenuis</i> , cat, HDM)	12 mo: 681/706; 96	No significant difference between groups of “any” IgE-mediated disease; incidence of sensitization to egg at 12 mo was lower in the intervention group [34/368 (9%) vs. 52/338 (15%); <i>P</i> = 0.02]; incidence of IgE-associated eczema was lower in the intervention group, although not significant [26/368 (7%) vs. 39/338 (12%); <i>P</i> = 0.06]
Palmer et al., Australia, 2013 (28)	n = 706; Same population as (29)	—	Clinical examination by physician to determine incidence of eczema, asthma, allergic rhinitis, and food allergy; SPT (cow milk, egg, wheat, tuna, peanut, cashew, sesame, grass pollen, perennial ryegrass, olive tree pollen, <i>A. tenuis</i> , cat, and HDM)	3 y: 638/706; 90	No significant difference between “any” IgE-mediated disease in the first 3 y of life [64/368 (17.3%) vs. 76/338 (22.6%); <i>P</i> = 0.11]; no significant difference between clinical outcomes of eczema, allergic rhinitis, or asthma with or without sensitization

¹GA, gestational age; HDM, house dust mite; LC-PUFA, long-chain PUFA; NR, not reported; RCT, randomized controlled trial; ref, reference; SPT, skin-prick test.

Exposure/intervention

Observation of maternal dietary n-3 PUFA intake during pregnancy in cohort studies was ascertained by the use of diet history questionnaires (DHQs) or food-frequency questionnaires (FFQs). FFQs were either self- or interviewer-administered by phone or face-to-face. The timing of administration of the questionnaire varied between the first (19), second (17), and third (13, 15, 17, 20, 22–24) trimesters or ad hoc time points throughout pregnancy relative to study enrollment (16, 18). All of the studies required the women to complete the FFQ retrospectively from 3 d (21) to 3 mo (12, 14, 19) postpartum. The intensity of questioning in FFQs varied from 35 to 360 items, and total dietary fish exposure varied between 83 g/wk and 46 g/d (Table 1).

The intervention in 4 of 5 RCTs consisted of fish-oil capsules. Capsules varied in composition and dose of n-3 LC-PUFAs, ranging from 900 to 3700 mg of total n-3 LC-PUFAs/d (26, 29–31). One RCT supplied women with 2 × 150-mg portions of farmed salmon to consume twice per week, equivalent to 3450 mg n-3 LC-PUFAs/wk (the control group continued their habitual low-fish diet) (27). Placebos used in the double-blind RCTs consisted of olive oil (30, 31), soy oil (25, 26), or vegetable oil (28, 29) that did not contain n-3 LC-PUFAs. One study had 2 control groups (olive oil and “no oil”) (31). The timing of the intervention commenced between 20 and 30 wk of gestation and ceased at delivery of the infant in 4 of the included unique RCTs (27, 29–31). One RCT continued supplementation of mothers for 3.5 mo into the postpartum period (25, 26).

Clinical outcomes

The clinical indication of allergic disease in the offspring in observational studies and RCTs was diverse. Study outcomes reported included the following: symptoms of wheeze, eczema,

and rhinitis by parent report with the use of the International Study of Asthma and Allergy in Childhood (ISAAC) questionnaire (validated for use in 6- to 7-y-old children) (13, 15, 16, 18, 19, 22, 23); nonvalidated health questionnaire (21); parent report of physician diagnosis of asthma or eczema (12, 14, 20); asthma medication data from a social insurance database (22) or patient registry (31); and clinical examination by nurse or physician (17, 25–30). Five of the included observational studies and 4 of the RCTs assessed atopy outcomes with the use of either total and specific serum IgE analysis (24, 32), SPT (13, 14, 25, 26, 28–30), or a combination of both outcomes (12, 27). Individual or multiple outcomes were reported as cumulative incidences or point prevalences (Tables 1 and 2). The ages of the offspring at assessment of study outcomes varied between 6 mo and 8 y in observational studies (Table 1) and between 6 mo and 3 y in RCTs (Table 2), with the exception of one registry linkage trial that reported history of asthma diagnosis at 16 y (31).

Quality of observational studies

All of the included observational studies were assessed for methodologic quality by using the Newcastle-Ottawa Scale (33). It includes a “star system” (*) in which a study is judged on 3 domains: representativeness of study group selection (4 items), comparability of groups (2 items), and ascertainment of either the exposure or outcome (3 items) (see **Table 3**). Some level of bias was evident in all of the cohort studies, predominantly with regard to the ascertainment of maternal exposure to n-3 LC-PUFAs and/or assessment of allergic diseases outcomes. FFQs and DHQs are applicable to large cohorts and provide information on a wide range of foods. However, there are many limitations of these instruments, including dietary misreporting, which leads to dietary misclassification of intake and/or portion sizes. The detection of modest nutrient associations when using

TABLE 3
Summary of risk of bias assessment of included observational studies¹

Study	Quality indicators from Newcastle-Ottawa Scale (34)							
	Selection				Comparability		Outcome	
	Representativeness of the exposed cohort	Selection of the nonexposed cohort	Ascertainment of exposure	Demonstration that outcome of interest was not present at start of study	Comparability of cohorts on the basis of the design or analysis	Assessment of outcome	Was follow-up long enough for outcomes to occur?	Adequacy of follow-up of cohorts
Maslova (20)	*	*	*	*	*	*	*	
Miyake (18)	*	*		*	*	*	*	
Leermakers (19)	*	*		*	*	*	*	
Nwaru (23)		*			*	*	*	
Lumia (22)		*			*	*	*	*
Nwaru (24)		*			*	*	*	
Jedrychowski (17)	*	*	*	*	*	*	*	*
Miyake (16)	*	*		*	*	*	*	
Willers (15)	*	*		*	*	*	*	
Chatzi (14)	*	*	*		*	*	*	*
Romieu (12)	*	*	*		*	*	*	*
Willers (13)	*	*		*	*	*	*	*
Sausenthaler (21)	*	*		*	*	*	*	*

¹Each study is judged on three domains receiving a “*” to signify quality of each item within representativeness of study group selection (4 items); comparability of groups (2 items); and ascertainment of either the exposure or outcome (3 items).

an FFQ for dietary assessment is unlikely (34, 35). This is especially true for reliable estimates of absolute amounts of dietary fats (36, 37). A number of studies did not capture fish type (e.g., “fatty fish”) (Table 1), which may be problematic because there is significant variation in the amount of n-3 LC-PUFA intake depending on the type of fish consumed. Dietary intake over the preceding period of 4 wk to 3 mo was documented retrospectively and may be subject to recall bias. With the exception of 2 studies (17, 20), maternal diet was assessed only once (between 12 and 40 wk of gestation), which may not be indicative of diet throughout the whole of pregnancy, as shown by one of the Japanese studies (18). These authors conducted dietary assessment at “any time” between the 5th and 39th week of pregnancy and reported that 30% of respondents had substantial changes in their diet in the previous month.

Substantial geographic variation existed between cohort studies in relation to total fish intake, fish subgroups, and the number of types of fish consumed (38). Decreased variability between categories of fish or n-3 PUFA intake was evident in 4 included studies from the Netherlands (15, 19, 23, 24), a country where there is generally low fish consumption. In one study, of the 2760 included women, just 7% consumed fish >1.5 times/wk (30 g/d) and only one woman ate fish daily (15). In contrast, the mean fish intake in 2 Japanese studies was 48 g/d, with low variability between fish-intake quartiles due to the generalized high fish consumption. The presence of allergic disease symptoms in the offspring were self-reported in the majority of studies that used the ISAAC questionnaire (or a “modified” version). The ISAAC is a standardized questionnaire and has been validated in children aged 6–7 and 13–14 y old; however, most children in the included cohort studies were younger (15–19, 21–24). The ability to differentiate IgE-mediated allergic disease was diminished because only 4 of the 13 publications assessed atopy in the child (SPT or serum IgE) (12–14, 24). Attrition bias was possible in a number of studies with follow-up rates <80% (13, 15, 16, 18–20, 22–24). When comparing population characteristics of participants and nonparticipants, participants tended to display healthier lifestyle habits, higher maternal and paternal educational levels, and higher income levels (18, 20). One study with 3 publications included only infants at risk of developing diabetes by the presence of human leukocyte antigen in cord blood (DIPP study). Children with type 1 diabetes have been reported to have a reduced incidence of asthma and allergic diseases compared with the general population; therefore, this genetic susceptibility may further decrease the generalizability of this study’s findings.

Quality of RCTs

Two review authors (KPB and MM) assessed the risk of bias for each trial using the criteria outlined in the Cochrane Handbook for Systematic Reviews of Interventions (39). A summary of the risk of bias associated with each RCT is shown in **Table 4**. Two trials lacked sufficient clarity when reporting concealment of the allocation sequence (25–27) and were therefore assessed as having an unclear risk of bias. Blinding of participants was endeavored by image-matching capsules in 4 double-blind RCTs (25, 26, 28–31). One of these trials included a “no supplement” group, which may have resulted in unblinding of participants and personnel (31). All of the trials that used fish-oil capsules reported side effects of “fishy burps,” potentially risking unblinding of participants; however, objective outcome assessment, such as SPT, reduced the risk of performance bias. One single-blind trial that supplied salmon portions to women in the intervention group and no salmon in the control group was assessed as having a high risk of bias because the participants could not be blinded (27). This trial also had a high attrition rate, with only 62% of the control group attending the 6-mo follow-up, which is well below the minimum follow-up considered acceptable for minimizing attrition bias ($\geq 80\%$) (27). The risk of bias from incomplete outcome data reporting was high in 2 other trials (26, 30). One trial had significantly higher attrition in the treatment group (15%) than in the control group (2%) due to side effects perceived to be associated with the study product (30). Another excluded 25 mothers (17%) from the analysis because they did not complete supplementation for the requested period (26). Such postrandomization exclusions and attrition can contribute to systematic loss to follow-up and increase the risk of bias in already small and underpowered samples. Other bias was unclear in one registry linkage study due to a lack of clarity with regard to reporting of standardization of outcome assessments (31). Data were ascertained by searching the Danish National Patient Registry, a mandatory national hospital discharge register for asthma and allergic disease–related coding. Although the long-term follow-up rate for this study is outstanding, the overall rate of diagnosed asthma is much lower than reported in other studies. It is unknown how the diagnosis of asthma was made and likely that there was an underrepresentation of milder cases treated outside of the Danish hospital system. Of the 5 included RCTs, 4 studies used SPT as an outcome measure to determine atopy status of the offspring (26, 27, 29, 30); however, definitions of sensitization were inconsistent. Two studies used the standard clinical definition of wheal size ≥ 3 mm as the diagnosis of sensitization (27, 29).

TABLE 4

Summary of risk of bias assessment for included RCTs¹

First author (ref)	Random-sequence generation	Allocation concealment	Blinding of participants/personnel	Blinding of outcome assessment	Incomplete outcome data	Selective reporting	Other bias
Dunstan (30)	L	L	L	L	H	L	U
Olsen (31)	L	L	H	U	L	L	U
Furuhjelm (26)	L	L	L	L	L	H	L
Furuhjelm (25)		Follow-up of (26)		L	L	H	L
Noakes (27)	L	H	H	L	H	L	H
Palmer (29)	L	L	L	L	L	L	L
Palmer (28)		Follow-up of (29)		L	L	L	L

¹H, high risk of bias; L, low risk of bias; RCT, randomized controlled trial; ref, reference; U, unclear risk of bias.

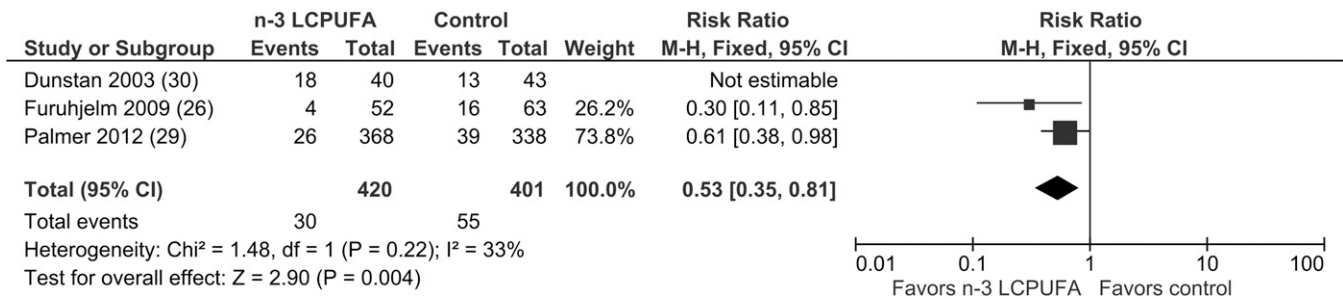


FIGURE 2 Forest plot of randomized controlled trials comparing maternal n-3 LC-PUFA supplementation during pregnancy with placebo on atopic eczema (eczema with positive skin prick test) in the offspring in the first 12 mo of life. In this fixed-effects model, squares represent RRs and error bars represent 95% CIs. The diamond represents the overall effect estimate. There was no significant heterogeneity between studies. The randomized controlled trial by Dunstan et al. (30) was omitted from this forest plot because confirmation of atopy (positive skin prick test) for eczema outcomes was not reported. LC-PUFA, long-chain PUFA; M-H, Mantel-Haenszel.

Another 2 studies that conducted 6- and 12-mo follow-ups of infants included results of ≥ 2 -mm wheal size in the analysis of sensitization (26, 30). The rationale for this criterion as opposed to the standard clinical definition was not reported. There is evidence that SPT reactions in the very young (<12 mo) are often diminished, which makes interpretation difficult (40).

Results of observational studies

Table 1 summarizes all of the identified studies that investigated the association between maternal n-3 LC-PUFA or fish intake and allergic disease outcomes in the offspring. Eight of the 13 included studies found a protective association between increased n-3 LC-PUFA or fish intake in the prenatal diet and incidence of one or more allergic disease symptoms in the child (12–14, 17, 18, 20–22). One of these studies assessed maternal diet by phone interview in addition to self-completed FFQ; however, a significant association of increased fish intake on asthma outcomes was only evident from phone interview data (20). Four studies showed no association between maternal n-3 LC-PUFA or fish intake and allergic disease outcomes (15, 16, 23, 24). One study reported an adverse association in one category of fish intake (19).

Eczema

The incidence of eczema was reported in 8 studies, with 4 of these showing that higher fish/n-3 LC-PUFA exposure was associated with a reduction in the risk of eczema in the offspring of between 27% and 43%. Fish intake of >205 g/wk (17) and fish intake 2.5 times/wk compared with no intake (12) were protective against the risk of eczema at 12 mo. High compared

with low fish intake was inversely associated with parent reported, doctor diagnosed eczema at 24 mo in one study (21) and fish intake of ≥ 1 time/wk compared with never was associated with a decreased risk of doctor-confirmed eczema and currently treated eczema (13). Two separate studies from Japan found no association between increased maternal fish intake and eczema (16, 18), and one of the publications from the DIPP study found no association between increased total n-3 PUFAs, DHA, or EPA with eczema (16, 18, 23). One study from the Netherlands reported that the midcategory of intake (35–69 g/wk) was associated with the highest eczema rate, whereas the lower and higher categories of fish intake (1–34 and >70 g/wk) showed no association with overall risk of eczema (19).

Rhino-conjunctivitis

Three studies assessed symptoms of rhino-conjunctivitis (13, 20, 23). One study concluded that maternal fish intake once or more per week compared with never was protective for “doctor-confirmed hay fever” at 5 y (13). Two studies found no significant association between no maternal fish intake compared with high-frequency intake and rhino-conjunctivitis (20) or between increased total n-3 PUFAs, DHA, or EPA and rhino-conjunctivitis (23).

Asthma/wheeze

Nine studies assessed the incidence of asthma or symptoms of wheeze. Low maternal fish consumption was inversely related to parent report of physician diagnosis of asthma and wheezing symptoms at 18 mo and 7 y (20), atopic wheeze (12), and persistent wheeze at 6.5 y (14). Two studies reported a protective association of an increased total maternal n-3 LC-PUFA (but not



FIGURE 3 Forest plot of randomized controlled trials comparing maternal n-3 LC-PUFA supplementation during pregnancy with placebo on any eczema (eczema with or without a positive skin prick test) in the offspring in the first 12 mo of life. In this fixed-effects model, squares represent RRs and error bars represent 95% CIs. The diamond represents the overall effect estimate. There was no heterogeneity between studies. LC-PUFA, long-chain PUFA; M-H, Mantel-Haenszel.

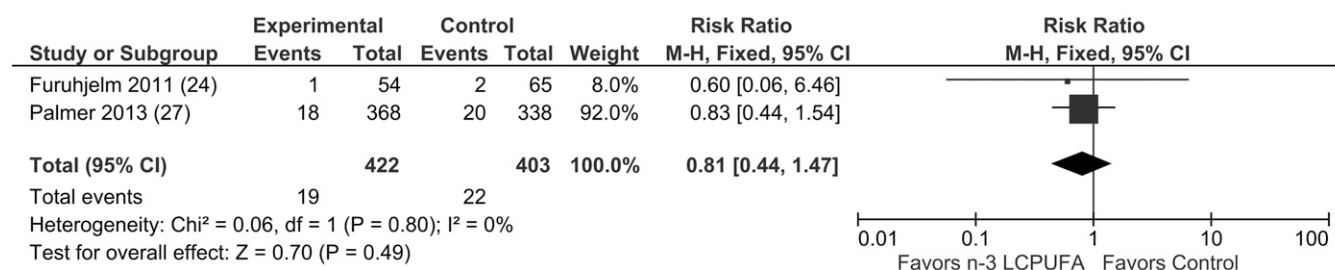


FIGURE 4 Forest plot of randomized controlled trials comparing maternal n-3 LC-PUFA supplementation during pregnancy with placebo on cumulative incidence of IgE-mediated rhino-conjunctivitis (rhino-conjunctivitis with a positive skin prick test) in the offspring within the first 3 y of life. In this fixed-effects model, squares represent RRs and error bars represent 95% CIs. The diamond represents the overall effect estimate. There was no heterogeneity between studies. LC-PUFA, long-chain PUFA; M-H, Mantel-Haenszel.

fish) intake on wheeze at 24 mo (18) and asthma at 5 y (22). One Japanese study showed that high maternal n-3 PUFA intake (via α -linoleic acid, 18:3n-3) was associated with a reduced risk of wheeze (16). The 3 remaining studies (2 from the Netherlands) found no significant association between increased fish or n-3 LC-PUFA consumption and wheeze (23), risk of asthma (22), or asthma symptoms (13).

Sensitization

Three studies reported the association of fish or n-3 PUFA intake on total and specific IgE at 2 y (21), 4 y (12), and 5 y (24), all showing no significant association. One of these studies conducted further follow-up at 6.5 y of age with SPT and reported that fish intake of ≥ 2.5 times/wk was protective against sensitization to house dust mite (12). One study that performed an SPT at 5 y of age found no significant association between maternal fish intake and atopic sensitization (13).

Results of RCTs

Table 2 summarizes all of the identified RCTs that investigated the effect of prenatal n-3 LC-PUFA supplementation (or supplied fish portions) on allergic disease outcomes in the offspring. Of the 7 publications (from 5 unique RCTs), 5 found a protective effect on ≥ 1 clinical outcomes of allergic disease or sensitization at ≥ 1 assessment time points.

Eczema

The incidence of IgE-mediated eczema between children born to mothers supplemented with n-3 LC-PUFAs during pregnancy and control groups was assessed between 0 and 3 y in 4 RCTs

and reported in 6 of the included publications (25–30). One trial that conducted follow-up of children over a 2-y period found a significant reduction in the cumulative incidence of atopic eczema at 12 and 24 mo (25, 26). Another study reported a non-significant protective effect of the intervention at 12 mo, with no effect when reassessed at 3 y (28, 29).

The 2 remaining studies found no effect of n-3 LC-PUFA supplementation on eczema (27, 30), although one did report a significant reduction in severity of disease (30). Results from RCTs that reported outcomes of “atopic eczema” (eczema symptoms and positive SPT) at 12 mo of age were combined in a meta-analysis showing a significant reduction in the incidence of atopic eczema (RR: 0.53; 95% CI: 0.35, 0.81; $P = 0.004$) (Figure 2). Both of these trials also reported outcomes of incidence of “any” eczema (eczema symptoms with or without positive SPT) at 12 mo, showing no significant effect of the intervention (RR: 0.85; 95% CI: 0.67, 1.07; $P = 0.16$) (Figure 3).

Rhino-conjunctivitis

There was no difference between the groups in the cumulative incidence of “any rhino-conjunctivitis” or “IgE-mediated rhino-conjunctivitis” in the 2 studies that reported this outcome at 0–24 mo (25) and 0–3 y (29) (RR: 0.81; 95% CI: 0.44, 1.47; $P = 0.49$) (Figure 4).

Asthma

Asthma and/or symptoms of asthma were assessed in 5 of the 7 included publications at different ages and with a variety of outcome measures, which prevented meaningful meta-analysis. There were no differences in asthma or wheeze with or without sensitization at 6 mo (27), 12 mo (30), 24 mo, 0–24 mo (25), or 3 y (28).



FIGURE 5 Forest plot of randomized controlled trials comparing maternal n-3 LC-PUFA supplementation during pregnancy with placebo on the incidence of a positive skin prick test result to any allergen extracts tested in the offspring within the first 12 mo of life. In this fixed-effects model, squares represent RRs and error bars represent 95% CIs. The diamond represents the overall effect estimate. There was no heterogeneity between studies. LC-PUFA, long-chain PUFA; M-H, Mantel-Haenszel.

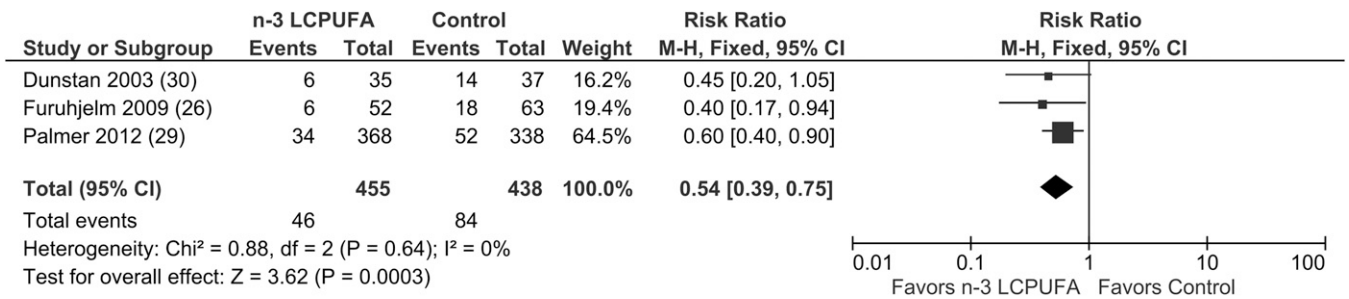


FIGURE 6 Forest plot of randomized controlled trials comparing maternal n-3 LC-PUFA supplementation during pregnancy with placebo on the incidence of a positive skin prick test to hen’s egg in the offspring in the first 12 mo of life. In this fixed-effects model, squares represent RRs and error bars represent 95% CIs. The diamond represents the overall effect estimate. There was no heterogeneity between studies. LC-PUFA, long-chain PUFA; M-H, Mantel-Haenszel.

One registry-based follow-up RCT included asthma diagnosis as an outcome and reported a significant reduction in “any asthma” and “allergic asthma” at 16 y of age in the fish-oil-supplemented group compared with the “olive oil” control group. However, a significant positive effect on “any asthma” and “allergic asthma” was also evident in the second “no oil” control group (31).

Sensitization

Sensitization to common allergens was assessed by SPT at 6 mo (27), 12 mo (26, 27, 29, 30), 24 mo (25), and 3 y (28). Three RCTs with sensitization outcomes at 12 mo of age were combined in meta-analysis and showed a significant protective effect of the intervention on the period prevalence of “any positive SPT” (RR: 0.68; 95% CI: 0.52, 0.89; *P* = 0.006) (**Figure 5**). The trial with the salmon portion-based intervention also measured total IgE at birth and 6 mo of age, with no difference between the groups (27). A meta-analysis of sensitization to individual allergen extracts showed a significant reduction in “sensitization to egg” at 0–12 mo (RR: 0.55; 95% CI: 0.39, 0.76; *P* = 0.0004) (**Figure 6**) and “sensitization to any food” at 12 mo (RR: 0.59; 95% CI: 0.46, 0.76; *P* < 0.0001) (**Figure 7**).

DISCUSSION

Our systematic review and meta-analysis shows that increased prenatal intakes of n-3 LC-PUFAs in observational studies and RCTs are suggestive of benefits. However, due to the inconsistency of results, the hypothesis linking increased maternal n-3 LC-PUFA intake to a protective association or effect on childhood allergic disease cannot unequivocally be confirmed or rejected. Despite the challenges of capturing and comparing

outcomes of a dynamic disease, some of the consistency observed across the study designs used is noteworthy (41). It is widely acknowledged that causal inferences based on results from studies of nutritional epidemiology that have been confirmed by results from RCTs represent more persuasive evidence than those elucidated from observational data alone (10). Observational studies showed a trend toward a protective association of increased maternal n-3 LC-PUFAs on the incidence of eczema and wheeze or asthma. Similar results were evident in RCTs for eczema but not for wheeze or asthma, perhaps due to the age of the child when outcome assessments were conducted (1–3 y). The most significant and consistent outcome in RCTs was a reduction in the incidence of sensitization (positive SPT) in the n-3 LC-PUFA group. Although the presence of an IgE-mediated reaction to a food or inhalant allergen generally confirms that an individual is “atopic,” the relation between sensitization and symptoms of allergic disease is known to be complex and dynamic. This finding may have some correlation to the reduction in respiratory disease evident in observational studies in school-aged children. It is well documented that early sensitization to food allergens (especially egg) is shown to be a valuable predictor of subsequent sensitization to aero-allergens and allergic respiratory diseases (42–45).

Heterogeneity between study designs may, in part, be explained by the limited number of well-powered high-quality RCTs with long-term follow-up and consistent age-appropriate outcomes. Whereas epidemiologic studies have the potential to assess long-term health effects of nutrient intake, the possibility for residual confounding cannot be discounted, particularly when assessing single-nutrient exposure (35). The protective associations seen in the offspring of women with increased intakes of n-3 LC-PUFAs

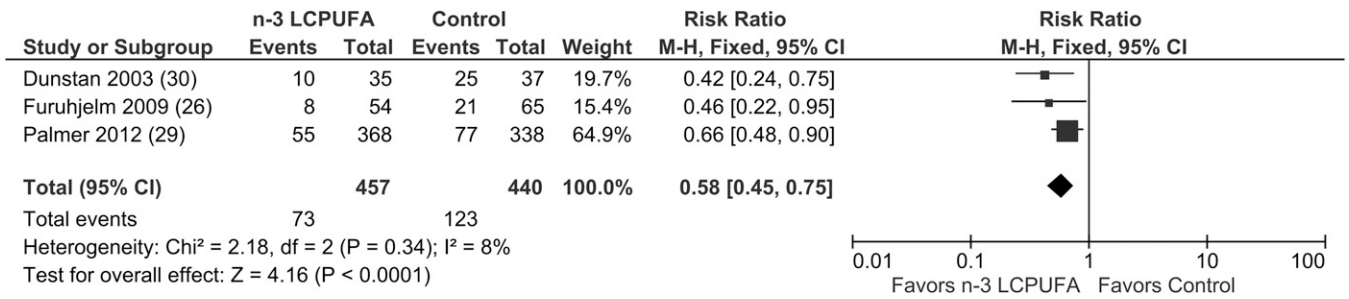


FIGURE 7 Forest plot of randomized controlled trials comparing maternal n-3 LC-PUFA supplementation during pregnancy with placebo on the incidence of a positive skin prick test to any food extract in the offspring in the first 12 mo of life. In this fixed-effects model, squares represent RRs and error bars represent 95% CIs. The diamond represents the overall effect estimate. There was no heterogeneity between studies. LC-PUFA, long-chain PUFA; M-H, Mantel-Haenszel.

during pregnancy may not be due to this exposure, but rather serve as a proxy indicator for a number of other maternal characteristics that are associated with the healthy development of their offspring (46). These confounding factors are difficult to fully accommodate in data analysis, even when acknowledged. In addition, dietary intake in epidemiologic studies is notoriously difficult to measure and often results in variations in findings (47). Flaws in standard tools for dietary assessment (FFQ or DHQ) are well documented and cannot be ignored (35, 37, 47, 48). This issue was emphasized in the large Danish birth cohort ($n = 28,936$) that observed an association between fish intake and asthma diagnosis (20). Self-completed FFQ data showed a null association between fish intake and asthma diagnosis at 18 mo and 7 y; however, analysis of fish-intake data collected by telephone interview showed a significant inverse relation (20).

The main strength of our integrated systematic review, which included both prospective observational studies and RCTs, lies in the detailed search strategy, rigorous and transparent methods, and the qualitative and quantitative synthesis of differing methodologies. However, the risk of publication bias cannot be excluded because only published studies were included in meta-analysis. Using well-established processes for conducting systematic reviews (49) we determined that observational studies and many RCTs had methodologic weaknesses, making it difficult to draw any strong inferences from this review. There have been numerous studies that investigated maternal dietary $n-3$ LC-PUFA status on the regulation of the fetal immune response (50–54). These studies reported plausible biological mechanisms that may modulate the development of allergic disease in infants with a genetic risk. However, as is evident in this and other reviews of increased maternal supply of $n-3$ LC-PUFAs and outcomes of allergy (5–9, 55–57), results are promising but inconsistent. Continued supplementation of breastfeeding mothers during the early postpartum period to compensate for the natural decline in maternal plasma $n-3$ LC-PUFAs may provide additional benefit to the development and maturation of the infant immune system and is worthy of further investigation (58). Further evidence from well-powered, high-quality trials of comparable methodology and standardized, objective outcome assessments are essential to investigate the optimal dose and tolerability and to conclusively establish benefits. Due to the dynamic nature of allergic disease, RCTs should continue follow-up of children throughout childhood, at least until school age to investigate if the significant protective effects of maternal $n-3$ LC-PUFA supplementation seen on eczema and sensitization in early life will affect the progression of the atopic march.

It is generally agreed that the increasing number of people affected by allergic disease is a consequence of our 21st-century lifestyle. Almost certainly this increase is attributable to genetic and environmental factors, including, but not limited to, diet. Diet is easily modifiable; perhaps this simple, low-cost approach of increasing $n-3$ LC-PUFA intake during pregnancy may offer the best opportunity for a primary prevention strategy, decreasing the burden of allergic disease for future generations.

The authors' responsibilities were as follows—KPB and MM: jointly conceived the systematic review; KPB: conducted searches, assessed inclusion, extracted data, assessed validity, performed meta-analyses, and wrote the manuscript; and all authors: critically reviewed the manuscript for content and approved the final version. MG and MM took part in 2 of the trials in the systematic review. MM is currently chief investigator in a large-scale clinical

trial of marine oil supplementation in the perinatal period; she has no financial interests in the production or sales of nutritional supplements or infant formula but has provided scientific advice to the nutritional supplement and formula industry. MM serves on the scientific advisory for Nestlé, Nutricia, and Fonterra. Associated honoraria for MM are paid to her institution to support conference travel and continuing education for postgraduate students and early-career researchers. None of the other authors declared a conflict of interest. There was no input from the funding bodies in any aspect of design, implementation, analysis, or interpretation of the data.

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